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      (FILE 'MEDLINE, CANCERLIT, EMBASE, BIOSIS, CAPLUS, BIOTECHDS' ENTERED AT
      17:59:27 ON 01 FEB 2005)
                  DEL HIS
L1          16230 S FACTOR VII
L2        2003895 S MUTA?
L3           1211 S L2 AND L1
L4         420934 S CLEAVAGE
L5            990 S ENYZM?
L6         421889 S L5 OR L4
L7             80 S L6 AND L3
L8           4853 S FURIN OR SKI-1
L9              7 S L8 AND L7
L10             4 DUP REM L9 (3 DUPLICATES REMOVED)
L11            39 DUP REM L7 (41 DUPLICATES REMOVED)
L12            18 S L1 AND L8
L13            12 DUP REM L12 (6 DUPLICATES REMOVED)

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pathway on the cell surface that ultimately leads to thrombin formation.

L11 ANSWER 31 OF 39 MEDLINE on STN DUPLICATE 10
AN 94264305 MEDLINE
DN PubMed ID: 8204879
TI Severe **factor VII** deficiency caused by **mutations** abolishing the **cleavage** site for activation and altering binding to tissue factor.
AU Chaing S; Clarke B; Sridhara S; Chu K; Friedman P; VanDusen W; Roberts H R; Blajchman M; Monroe D M; High K A
CS Department of Medicine, University of North Carolina at Chapel Hill.
NC K08-HL01922 (NHLBI)
P01-HL06350 (NHLBI)
SO Blood, (1994 Jun 15) 83 (12) 3524-35.
Journal code: 7603509. ISSN: 0006-4971.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Abridged Index Medicus Journals; Priority Journals
EM 199407
ED Entered STN: 19940721
Last Updated on STN: 19940721
Entered Medline: 19940712
AB **Factor VII** (F.VII) is a vitamin-K-dependent serine protease required in the early stages of blood coagulation. We describe here a patient with severe F.VII deficiency, with a normal plasma F.VII antigen level (452 ng/mL) and F.VII activity less than 1%, who is homozygous for two defects: a G-->A transition at nucleotide 6055 in exon 4, which results in an Arg-->Gln change at amino acid 79 (R79Q); and a G-->A transition at nucleotide 8961 in exon 6, which results in an Arg-->Gln substitution at amino acid 152 (R152Q). The R79Q **mutation** occurs in the first epidermal growth factor (EGF)-like domain, which has previously been implicated in binding to tissue factor. The R152Q **mutation** occurs at a site (Arg 152-Ile 153) that is normally cleaved to generate activated F.VII (F.VIIa). Analysis of purified F.VII from patient plasma shows that the material cannot be activated by F.Xa and cofactors. In addition, in an in vitro binding assay using relipidated recombinant tissue factor, patient plasma showed markedly reduced binding to tissue factor at all concentrations tested. In an effort to separate the contributions of the two **mutations**, three recombinant variants, wild-type, R79Q, and R152Q, were prepared and analyzed. The R152Q variant had markedly reduced activity in a clotting assay, whereas R79Q showed a milder, concentration-dependent reduction. The R152Q variant exhibited nearly normal binding in the tissue factor binding assay, whereas the R79Q variant had markedly reduced binding. The time course of activation of the R79Q variant was slowed compared with wild-type. Our results suggest that the first EGF-like domain is required for binding to tissue factor and that the F.VII zymogen lacks activity and requires activation for expression of biologic activity.

L11 ANSWER 18 OF 39 BIOSIS COPYRIGHT (c) 2005 The Thomson Corporation. on STN

AN 2001:311593 BIOSIS

DN PREV200100311593

TI A novel **mutation** in **factor VII** (V154G: FVIIPhiladelphia) impairs tissue factor binding and FVII coagulant activity.

AU Toso, Raffaella [Reprint author]; Tidd, Theresa [Reprint author]; High, Katherine A. [Reprint author]; Pinotti, Mirko [Reprint author]; Marchetti, Giovanna; Castaman, Giancarlo; Bernardi, Francesco; Pollak, Eleanor S. [Reprint author]

CS Research Hematology, Children's Hospital of Philadelphia, Philadelphia, PA, USA

SO Blood, (November 16, 2000) Vol. 96, No. 11 Part 1, pp. 260a. print. Meeting Info.: 42nd Annual Meeting of the American Society of Hematology. San Francisco, California, USA. December 01-05, 2000. American Society of Hematology.
CODEN: BLOOAW. ISSN: 0006-4971.

DT Conference; (Meeting)
Conference; Abstract; (Meeting Abstract)
Conference; (Meeting Poster)

LA English

ED Entered STN: 27 Jun 2001
Last Updated on STN: 19 Feb 2002

AB **Factor VII** is a 406 amino acid single chain vitamin K-dependent protein that circulates in blood as a zymogen and is cleaved at amino acid 152 into a two-chain serine protease, FVIIa. When complexed with the integral membrane protein Tissue Factor (TF), TF/FVIIa plays a pivotal role in initiating blood coagulation. We describe here a novel FVII **mutation** (FVIIPhiladelphia) located in exon 6 (GTG to GGG nt. 8967) that causes substitution of a Val for a Gly at amino acid 154. Genotypes and plasma FVII levels of 3 carriers of FVII-V154G from USA and Italy are shown. Recombinant FVII-V154G was expressed in HEK 293 cells and purified using a Ca2+-dependent immuno-affinity column. The FVII-V154G **mutation** (P2' residue, chymotrypsin numbering 17), 2 amino acids away from the activation site at 152, does not impair conversion of zymogen protein to the two-chain active form as activated by human FIXa, FXa, FXIIa or the complex soluble TF/FVIIa. FVIIa-V154G has no activity toward the macromolecular substrates FVII, FIX or FX, but retains 2% activity toward a FVIIa chromogenic substrate (Spec VIIa). In order to assess FVII/FVIIa binding to tissue factor, two recombinant FVII **mutants** were expressed, one (R152Q) resistant to activation and the other (S344A) with no FVIIa activity after **cleavage**. The zymogen FVII R152Q and FVIIa-V154G showed a 20 to 40-fold decreased affinity for two different sources of soluble TF as compared to the S344A FVIIa **mutant** but only a 5 fold difference using relipidated TF. Comparison of FVIIa-V154G with FVIIa and other serine protease models show that FVIIa-V154G is unable to form the critical salt-bridge; these findings imply that **cleavage** of the zymogen FVII to the two-chain form must be accompanied by the formation of the salt-bridge to enable activity of the TF-FVIIa complex. In summary, modification of the P2' amino acid found in FVII-V154G does not prevent **cleavage** of zymogen FVII at the 152 activation site but promotes binding characteristics similar to zymogen FVII/TF.

L11 ANSWER 16 OF 39 MEDLINE on STN DUPLICATE 5
AN 2001124906 MEDLINE
DN PubMed ID: 11139238
TI **Factor VII** deficiency and the FVII **mutation** database.
AU McVey J H; Boswell E; Mumford A D; Kemball-Cook G; Tuddenham E G
CS MRC Clinical Sciences Centre, Imperial College School of Medicine, London, UK.. john.mcvey@csc.mrc.ac.uk
SO Human mutation, (2001) 17 (1) 3-17. Ref: 81
Journal code: 9215429. ISSN: 1098-1004.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
General Review; (REVIEW)
(REVIEW, TUTORIAL)
LA English
FS Priority Journals
EM 200102
ED Entered STN: 20010322
Last Updated on STN: 20021211
Entered Medline: 20010222
AB **Factor VII** (FVII) is a zymogen for a vitamin K-dependent serine protease essential for the initiation of blood coagulation. It is synthesized primarily in the liver and circulates in plasma at a concentration of approximately 0.5 microg/ml (10 nmol/L). The FVII gene (F7) is located on chromosome 13 (13q34), consists of 9 exons, and spans approximately 12kb. It encodes a mature protein of 406 amino acids, which has an N-terminal domain (Gla) post-translationally modified by gamma-carboxylation of glutamic acid residues, two domains with homology to epidermal growth factor (EGF1 and 2), and a C-terminal serine protease domain. The single chain zymogen is activated by proteolytic **cleavage** at Arg152-Ile153. There are 238 individuals described in the world literature with **mutations** in their F7 genes (FVII **mutation** database; europium.csc. mrc.ac.uk). Complete absence of FVII activity in plasma is usually incompatible with life, and individuals die shortly after birth due to severe hemorrhage. The majority of individuals with **mutations** in their F7 gene(s), however, are either asymptomatic or the clinical phenotype is unknown. In general, a severe bleeding phenotype is only observed in individuals homozygous for a **mutation** in their F7 genes with FVII activities (FVII:C) below 2% of normal, however, a considerable proportion of individuals with a mild-moderate bleeding phenotype have similar FVII:C by in vitro assay. The failure of in vitro tests to differentiate between these groups may be due to lack of sensitivity in the assays to the very low amounts of FVII:C, which are sufficient to initiate coagulation in vivo. A number of polymorphisms have been identified in the F7 gene and some have been shown to influence plasma FVII antigen levels.

L13 ANSWER 4 OF 12 CAPLUS COPYRIGHT 2005 ACS on STN
AN 2001:713370 CAPLUS
DN 135:277991
TI Modified blood clotting factors for treatment of bleeding or clotting disorder
IN High, Katherine A.; Margaritis, Paris; Camire, Rodney M.
PA Children's Hospital of Philadelphia, USA
SO PCT Int. Appl., 55 pp.
CODEN: PIXXD2
DT Patent
LA English
FAN.CNT 1

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	WO 2001070763	A1	20010927	WO 2001-US9355	20010322
	W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
	RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG				
	US 2004102388	A1	20040527	US 2001-816688	20010322
PRAI	US 2000-191331P	P	20000322		

AB The invention provides compns. including modified blood clotting factors, i.e., **Factor VII**, Factor IX, and Factor X, that have a non-native proteolytic cleavage site engineered into them allowing intracellular cleavage and secretion of an active form. The compns. are useful in the methods for treating a bleeding or clotting disorder. For example, gene transfer of modified blood coagulation factor VIIa using the AAV-hAAT-ApoE-FVIIa expression vector offers a treatment for hemophilia patients and does not appear to induce production of inhibitory antibodies against FVIIa.